Attorney Docket No.: 5600.200-US

PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of: Sandal et al

Confirmation No: 1759

Serial No.: 09/426,340

Group Art Unit: 1655

Filed: October 25, 1999

Examiner: Johannsen, D.

For: Method For Generating A Gene Library

VERSION WITH MARKINGS TO SHOW CHANGES MADE

Sir:

Below is a marked-up version of the amendments made in the accompanying amendment.

IN THE CLAIMS:

Claims 1 and 21 have been amended as follows:

1. (Three-times Amended) A method for generating a gene library from an environmental pool of organisms isolated from soil, animal dung, insect dung, insect gut, animal stomach, sea or lake water, waste water, sludge, or sediment, which gene library is enriched in DNA encoding a polypeptide with an activity of interest, which method comprises:

a) subjecting the environmental pool of organisms to cultivation under conditions wherein the pool of organisms is enriched in organisms harbouring said DNA, wherein said enriched pool of organisms is prepared without screening the organisms for presence of the activity of interest; and

b) preparing a gene library [directly] from the enriched environmental pool of organisms without screening the organisms for presence of the activity of interest.

Accordingly, Applicants submit that the claims overcome this rejection under 35 U.S.C. 103. Applicants respectfully request reconsideration and withdrawal of the rejection.

2. (Unchanged) The method of claim 1, wherein the conditions are culturing in a medium that contains a substrate for the gene product encoded by said DNA.

3. (Unchanged.) The method of claim 2, wherein the substrate constitutes the carbon source and/or nitrogen source of the medium.

4. (Unchanged.) The method of claim 2, wherein the substrate comprises pectin, amylose, cellulose, galactose, xylose or arabinose or a combination thereof.

5. (Unchanged.) The method of claim 1, wherein the pool of organisms is enriched by one or more growth restrictions.

6. (Unchanged.) The method of claim 5, wherein the growth restrictions comprise pH and temperature.

7. (Unchanged.) The method of claim 5, wherein the growth restrictions are pH 9-11 and temperature 50-70°C.

8. (Unchanged.) The method of claim 1, wherein the environmental pool of organisms is isolated from an animal stomach or an insect gut.

9. (Unchanged.) The method of claim 8, wherein the pool of organisms is isolated from a cow's rumen.

10. (Unchanged.) The method of claim 8, wherein the pool of organisms is isolated from the gut of an insect of the *Isoptera*, *Lepidoptera*, *Coleoptera*, or *Diptera* families.

11. (Unchanged.) The method of claim 10, wherein the pool of organisms is isolated from the gut of insects selected from the group consisting of *Agrotis, Neotermes castaneus, Tineola bisselliella*, and *Melolontha vulgaris.*

12. (Unchanged.) The method of claim 8, wherein prior to isolation, the pool of organisms is enriched by supplying feed to the animal or insect, which comprises a substrate for the polypeptide with an activity of interest.

13. (Unchanged) The method of claim 1, wherein the gene library is enriched in DNA encoding an enzyme of interest.

14. (Unchanged.) The method of claim 13, wherein the enzyme of interest comprises a hydrolase, an oxidoreductase, a transferase, a lyase or a ligase.

15. (Unchanged.) The method of claim 14, wherein the enzyme of interest comprises a protease, lipase, beta-galactosidase, lactase, polygalacturonase, beta-glucoamylase, esterase, hemicellulase, peroxidase, oxidase, laccase or glucose oxidase.

16. (Unchanged.) The method of claim 14, wherein the enzyme of interest is a pectinase, an amylase, a galactanase, an arabinase, a xylanase, or a cellulase.

17. (Unchanged.) The method of claim 1, wherein the environmental pool of organisms comprises microorganisms.

18. (Unchanged.) The method of claim 17, wherein the environmental pool of organisms comprises enzyme producing microorganisms.

19. (Unchanged.) The method of claim 17, wherein the microorganisms comprise *Eubacteria*, *Archaebacteria*, fungi, algae and/or protozoa.

21. (Three-times Amended) A method of [selecting] identifying a DNA sequence encoding a polypeptide of interest from an environmental pool of organisms isolated from soil, animal dung, insect dung, insect gut, animal stomach, sea or lake water, waste water, sludge, or sediment, which method comprises:

a) subjecting the environmental pool of organisms to cultivation under conditions wherein the pool of organisms is enriched in organisms harbouring said DNA, wherein said enriched pool of organisms is prepared without screening the organisms for presence of the activity of interest;

b) producing gene libraries [directly] from the enriched environmental pool of organisms without screening the organisms for presence of the activity of interest; and

c) screening the libraries of step b) [for] to identify a DNA encoding the polypeptide of interest.

22. (Unchanged.) A method of claim 21, wherein the polypeptide of interest encodes an enzyme.

23. (Unchanged.) The method of claim 21, wherein the gene libraries are screened in step c) for an active enzyme.

24. (Unchanged) The method of claim 21, wherein the polypeptide of interest encodes one of a pectinase, amylase, galactanase, arabinase, xylanase or cellulase.

25. (Unchanged.) A gene library prepared from an environmental pool of organisms enriched in DNA encoding a polypeptide with an activity of interest by the method of claim 1.

27. (Unchanged.) The gene library of claim 25, wherein the polypeptide is an enzyme which comprises a pectinase, an amylase, a galactanase, an arabinase, a xylanase or a cellulase.